Sudden cardiac death: **A matter of faulty ion channels?**

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Recent evidence suggests that various forms of sudden cardiac death in people with hearts that apparently function normally are caused by inherited or *de novo* mutations in genes coding for ion channel subunits.

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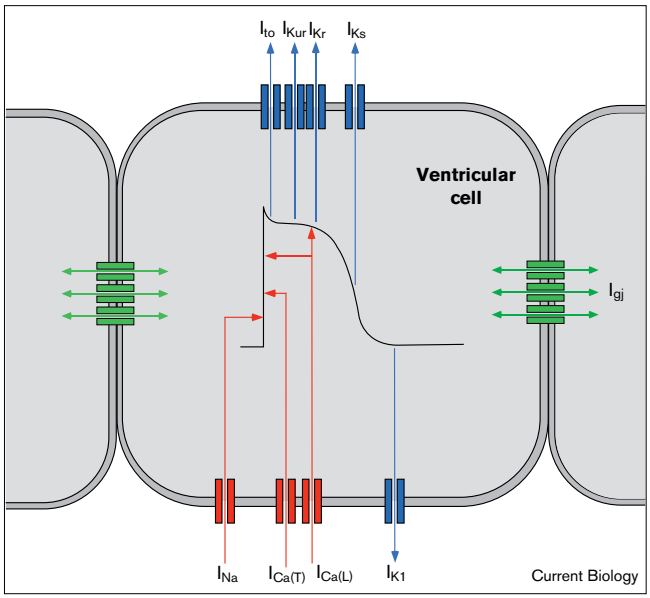
Coordinated contraction of the heart, which is essential for adequate blood supply to all parts of the body, is triggered by synchronized electrical activation of working myocardial cells. Upon stimulation, each cardiac myocyte generates its own electrical activation, or action potential, by virtue of the ion-conducting channels in their cell membrane. The ion channels act in an orchestrated manner to give a controlled cellular contraction. New insights into their importance have come from recent studies showing how inherited channel defects underlie some cases of sudden death through cardiac failure in people whose hearts seemed to be functioning normally.

The ions channels in the membranes of cardiac myocytes open and close in a time-dependent and voltage-dependent manner. Under resting conditions, most of the open channels are those selective for potassium ions; this keeps the membrane resting potential close to the potassium equilibrium potential, at about –80 mV. Activation occurs when the membrane is depolarized to values less negative than –60 mV. Sodium ion channels then open transiently, bringing the membrane potential to a value of about +30 mV, close to the sodium equilibrium potential. This regenerative process is rapidly terminated, because depolarization itself inactivates sodium channels. The membrane potential does not revert immediately to its resting value, because the initial depolarization also opens calcium channels, which activate and inactivate more slowly than sodium channels. Resumption of the resting potential depends on potassium channels, including ones that differ from those that maintain the membrane resting potential. Only when enough potassium channels have opened is the balance between inward (calcium) current and outward (potassium) current shifted in the net outward direction and repolarization ensues.

In recent years, the membrane currents that contribute to the generation of the ventricular action potential have been described in considerable detail (see Figure 1), and in some cases the genes coding for the protein subunits of the respective channels have been cloned and expressed in heterologous cell systems (Table 1). Unlike sodium and calcium channels, where the pore is formed by one large molecule consisting of four domains of six membrane-spanning α helices linked by intracellular loops, potassium channels are made from four molecules, each consisting of six membrane-spanning α helices. As cardiac ventricular cells contain mRNAs for many different types of potassium channel subunit [1], theoretically a large number of different homomeric or heteromeric channels are possible. This heterogeneity is likely to explain the considerable variation observed in ventricular action potential shape and duration, though rigorous proof of this is at present lacking.

In addition to these membrane currents, all cardiac myocytes are electrically coupled by gap junction channels [2], which allow current to flow between adjacent cells. This coupling means that action potentials generated in one cell depolarize adjacent cells, which subsequently also generate action potentials. In this way,

Figure 1



Simplified scheme of a ventricular cell. Each cell is connected to its neighbours by gap junction channels (green), through which current flows to depolarize the cell. This causes the activation of inward currents (red). I_{Na} and $I_{Ca(T)}$ inactivate rapidly, but $I_{Ca(L)}$ is sustained for longer. The depolarizing action of $I_{Ca(L)}$ is offset by successive activation of outward potassium currents (blue), resulting in a plateau phase of the action potential. When I_{Kr} and I_{Ks} are sufficiently activated, repolarization ensues and the membrane potential reverts to its resting value of about –85 mV; this level is kept stable by the action of I_{K1}.

Table 1

Currents that contribute to the cardiac action potential.

Current type	Ion carried	Current name	Probable channel subunit
Sodium current	Na^+	I_{Na}	SCN5A
Long-lasting calcium current	Ca^{2+}	$I_{Ca(L)}$	CACNLIA1
Transient calcium current	Ca^{2+}	$I_{Ca(T)}$	–
Transient outward current (4-aminopyridine-sensitive)	K^+	I_{to1}	Kv1.2, Kv1.4 Kv1.5, Kv2.1 Kv4.2/4.3
Transient outward current (calcium-activated)	K^+	I_{to2}	–
Ultrarapid delayed rectifier	K^+	I_{Kur}	Kv1.5
Rapid delayed rectifier	K^+	I_{Kr}	HERG
Slow delayed rectifier	K^+	I_{Ks}	KvLQT1 and KCNE1
Inward rectifier	K^+	I_{K1}	Kir2

This list of ion currents contributing to the ventricular action potential and the corresponding channel proteins is far from complete. Anionic current channels, and channels that operate only under abnormal conditions, have been omitted. Also, pumps and exhangers, such as the Na^+–Ca^{2+} exchanger, have been left out.

activation spreads from the sinoatrial node, the pacemaker of the heart where action potentials arise spontaneously, via the atria, the atrioventricular node and the ventricular conduction system towards both the right and the left ventricle. Under normal circumstances, action potentials are unidirectionally propagated in the heart for several reasons. Firstly, cardiac myocytes have a refractory period, which means that, once depolarized, they can only be activated again when their membrane potential has repolarized sufficiently to remove sodium and, to some extent, calcium channel inactivation. Secondly, the steep rate of rise of the action potential, together with the low intercellular resistance that results from the numerous gap junction channels between myocytes, ensures a high conduction velocity; retrogradely-conducted action potentials will therefore be blocked by tissue still in its refractory period. And thirdly, the morphology of the heart, and especially the ventricular conduction system, ensures an orderly activation sequence.

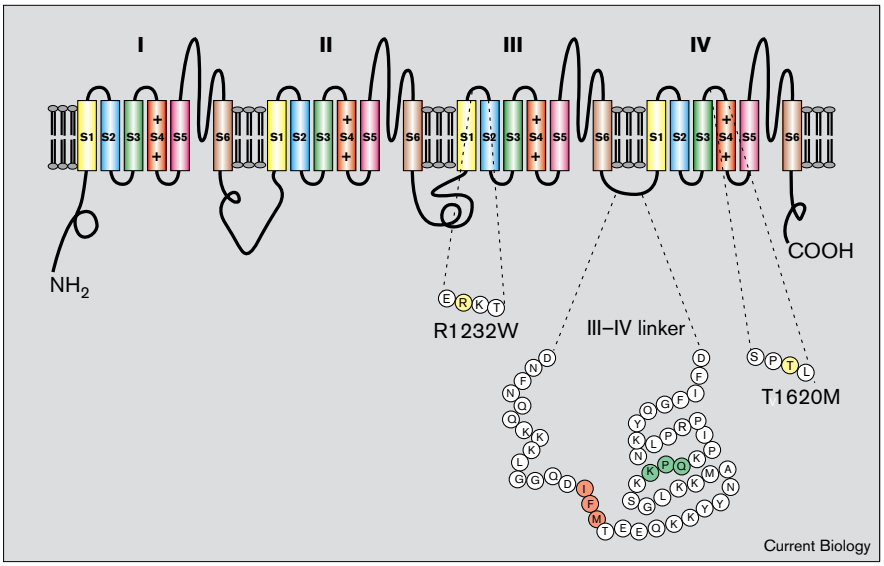
Cardiac arrhythmias may arise when any of the parameters mentioned above is pathologically changed. A lethal arrhythmia is actually the immediate cause of death in about 50% of the deaths due to cardiac disease. In most cases, such as heart failure, myocardial infarction or hypertrophic cardiomyopathy, structural changes set the scene for abnormal excitation or conduction, which under adverse conditions lead to arrhythmias. Less common are patients who suffer from ventricular arrhythmias without any sign of structural changes in the heart. Such arrhythmias may occur without any warning, deteriorate into ventricular fibrillation and lead to 'sudden cardiac death'. The mechanisms underlying these so-called primary electrical diseases are now beginning to be understood.

The first clues about the nature of primary electrical diseases came from the work on the inherited form of the so-called 'long QT' syndrome [3–5]. This relatively rare syndrome is characterised by an abnormally long 'QT interval' in the electrocardiogram. This is caused by a prolonged plateau phase of the ventricular action potential, which favors occurrence of the so-called 'early after-depolarizations' that result from reactivation of L-type calcium channels. Early after-depolarizations may trigger ventricular arrhythmias of the 'torsade de pointes' type, which under adverse conditions may degenerate into ventricular fibrillation.

Genetic analysis has linked at least five loci to the disease in various families with long QT syndrome, and four of the genes have been identified. These are: *SCN5A*, which encodes the cardiac sodium channel α subunit; *HERG*, which encodes a subunit of the potassium channel that carries the current I_{Kr}; and *KVLQT1* and *KCNE1*, which encode the α and β subunits, respectively, of the potassium channel carrying the current I_{Ks}. Mutations in *SCN5A*, notably one that causes a three amino-acid deletion in the linker between domains III and IV (Figure 2) that is involved in channel inactivation, lead to a phenotype in which there is a small but persistent tetrodotoxin-sensitive inward current at –10 to –20 mV, caused by reopening of inactivated channels [6]. Although the mechanism is not completely resolved, it seems likely that the deletion destabilizes the inactivated state. The consequence is that the balance between inward and outward currents that determines plateau phase duration is shifted in the inward direction, thereby prolonging it.

The mutations in *HERG*, *KVLQT1* and *KCNE1* frequently cause single amino-acid substitutions in their respective protein products. When expressed in heterologous cell systems, the mutant proteins were found not to form functional potassium channels, and they had a dominant-negative inhibitory effect on their corresponding wild-type proteins — when mutant and wild-type proteins were coexpressed, the potassium current density was less than expected from stoichiometric considerations [7,8]. How this dominant-negative effect is exerted is not clear; the kinetic properties of the current generally appeared normal. One possibility is that mutant and wild-type

Figure 2



Primary structure of the potassium channel protein SCN5A, and inferred membrane topology. The molecule has four domains, I–IV, each consisting of six membrane-spanning segments linked by intracellular and extracellular loops. S4 segments are voltage sensing elements responsible for the channel's voltage dependency. The so-called P-loops, which connect the S5 and S6 transmembrane helices, form the pore of the channel. The amino-acid sequence I-F-M (red) in the linker between domains III and IV is important for rapid inactivation of the channel. Deletion of amino acids KPQ (green) is responsible for one form of inherited long QT syndrome, characterised by incomplete inactivation. The combination of missense mutations R1232W and T1620M (yellow) has been implicated in one form of idiopathic ventricular fibrillation [10].

subunits combine to form a tetramer that does not integrate into the cell membrane.

Support for this possibility comes from recent work of London *et al.* [9], who have shown that overexpression in mouse heart of an amino-terminal fragment of Kv1.1, which normally forms a rat heart delayed-rectifier potassium channel, causes a prolonged QT interval and reduced levels of the channel protein Kv1.5, which normally contributes to repolarization. The truncated Kv1.1 had earlier been shown to have a dominant-negative effect on Kv1.5 in *Xenopus* oocytes, and to trap Kv1.5 subunits in the endoplasmic reticulum in GH3 cells. The implication is that mutant potassium channel subunits can combine with wild-type subunits to form abnormal channels that fail to reach the cell membrane. Whatever the precise mechanism, the consequence is that, in patients with potassium channel mutations, the balance between inward and outward currents during the plateau phase of the ventricular action potential is shifted in the inward direction, which leads to a prolongation of action potential duration and an increased risk of developing early afterdepolarizations.

Another primary electrical disease is idiopathic ventricular fibrillation where, without any previous warning, patients develop ventricular fibrillation and often die suddenly. Chen *et al.* [10] have recently found that mutations in *SCN5A* are carried in a few small families in which one member has been resuscitated from idiopathic ventricular fibrillation. These include a frame-shift mutation causing a premature stop codon in one family; a two-nucleotide insertion that disrupts an intronic splice-donor sequence in another family; and two missense mutations in a third family (see Figure 2). When expressed in oocytes, this last, doubly-mutated gene produced a sodium channel in which the steady-state inactivation potential was shifted 10 mV in the depolarizing direction, and a somewhat faster recovery from inactivation.

The mutations associated with idiopathic ventricular fibrillation thus appear to have a quite different effect from those found in long QT syndrome patients. It is, however, difficult to explain the phenotype on the basis of the observations with transfected oocytes. Electrocardiographic data show that the epicardial ventricular action potentials are shorter in these patients than normal, which leads to depolarizing current flow from endocardium to epicardium, especially in the relatively thin right ventricular wall. This may give rise to a premature beat which could initiate ventricular fibrillation. The suggestion is that the change in inactivation kinetics leads to a somewhat less intense inward sodium current; this in turn causes a slight reduction in L-type calcium channel activation and thus a decrease in inward current during the plateau phase, which leads to action potential shortening. The altered inactivation kinetics reported by Chen *et al.* [10] are, however, difficult to reconcile with this sequence of events.

Clearly, much more work is needed to resolve this matter. Mathematical models of ventricular action potentials are likely to be required to understand the complex interactions between the various ionic currents that contribute to the cardiac action potential, and the effects that small changes in channel kinetics have on it. Important new data are likely to be obtained in the next few years which ultimately will help to prevent sudden cardiac death,

which at present is responsible for over 300,000 deaths per year in the US alone.

References

1. Brahmajoti MV, Morales MJ, Liu S, Rasmusson RL, Campbell DL, Strauss HC: **In situ hybridisation reveals extensive diversity of K^+ channel mRNA in isolated ferret cardiac myocytes.** *Circ Res* 1996, **78:**1083-1089.
2. Gros DB, Jongsma HJ: **Connexins in mammalian heart function.** *BioEssays* 1996, **18:**719-730.
3. Wang Q, Shen J, Splawski I, Atkinson D, Li Z, Robinson JL, Moss AJ, Towbin JA, Keating MT: ***SCN5A* mutations associated with an inherited cardiac arrhythmia, long QT syndrome.** *Cell* 1995, **80:**805-811.
4. Wang Q, Curran ME, Splawski I, Burn TC, Milholland JM, VanRaay TJ, Shen J, Timothy KW, Vincent GM, de Jager T, *et al.*: **Positional cloning of a novel potassium channel gene: *KVLQT1* mutations cause cardiac arrhythmias.** *Nat Genet* 1996, **12:**17-23.
5. Sanguinetti MC, Jiang C, Curran ME, Keating MT: **A mechanistic link between an inherited and an acquired cardiac arrhythmia: HERG encodes the I_{Kr} potassium channel.** *Cell* 1995, **81:**299-307.
6. Chandra R, Starmer CF, Grant AO: **Multiple effects of *KPQ* deletion mutation on gating of human cardiac Na^+ channels expressed in mammalian cells.** *Am J Physiol* 1998, **274:**H1643-H1654.
7. Sanguinetti MC, Curran ME, Spector PS, Keating MT: **Spectrum of HERG K^+ channel dysfunction in an inherited cardiac arrhythmia.** *Proc Natl Acad Sci USA* 1996, **93:**2208-2212.
8. Shalaby FY, Levesque PC, Yang WP, Little WA, Conder ML, Jenkins-West T, Blanar MA: **Dominant-negative *KvLQT1* mutations underlie the LQT1 form of long QT syndrome.** *Circulation* 1997, **96:**1733-1736.
9. London B, Jeron A, Zhou J, Buckett P, Han X, Mitchell GF, Koren G: **Long QT and ventricular arrhythmias in transgenic mice expressing the N terminus and first transmembrane segment of a voltage-gated potassium channel.** *Proc Natl Acad Sci USA* 1998, **95:**2926-2931.
10. Chen Q, Kirsch GE, Zhang D, Brugada R, Brugada P, Brugada J, Potenza D, Moya A, Borggrefe M, Breithardt G, *et al.*: **Genetic basis and molecular mechanism for idiopathic ventricular fibrillation.** *Nature* 1998, **392:**293-296.